Mini-review: diabetic renal complications, a potential stinky remedy

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Submitted 6 July 2015; accepted in final form 2 November 2015

Sen U, Pushpakumar S. Mini-review: diabetic renal complications, a potential stinky remedy. Am J Physiol Renal Physiol 310: F119–F122, 2016. First published November 4, 2015; doi:10.1152/ajprenal.00299.2015.—Chronic kidney disease is one of the leading causes of end-stage renal disease (ESRD) throughout the world. Despite advances in understanding the disease process, the precise mechanism of initiation and progression remains unclear. In addition to high glucose, diabetes is associated with other metabolic derangements including protein (9, 30), lipid (35) and gaseous molecules such as, nitric oxide (NO) (41) and hydrogen sulfide (H2S) (45). Current evidence suggests that the pathogenesis of DN is multifactorial, and hyperglycemia mediates injury by several mechanisms such as fructokinase activation and ATP depletion, oxidative stress, production of inflammatory cytokines, activation of fibroblasts, and microaneurysm formation (12, 19, 36). Furthermore, at the cellular and molecular level an imbalance of matrix metalloproteinases and their inhibitors leads to abnormal extracellular matrix (ECM) metabolism (8, 27), and disrupted gap junction proteins cause poor cell-cell communication (43). A deficiency of nitric oxide has been implicated in advanced diabetic nephropathy (34). In recent studies, reduction of H2S-producing enzymes and plasma H2S has been associated with chronic kidney disease and diabetic nephropathy (2, 49). In light of current literature, this mini-review will focus on H2S biology and its role as a modulator and potential therapeutic agent in DN.

H2S Production in the Diabetic Kidney

In the last two decades, H2S has overcome its past reputation as a toxic gas and gained attention as a molecule involved in several biological functions. H2S was initially described as a neuromodulator by Abe and Kimura in 1996 (1). In the last decade, other studies have described its role in vasorelaxation (48), angiogenesis (4), nociception (28, 42), cytoprotection (15, 40), myocardial ischemia-reperfusion injury (7), atherosclerosis (5), including diabetic complications (25, 29). Endogenously, H2S is generated by cystathionine β-synthase (CBS), cystathionine γ-lyase (CSE), 3-mercaptopropionate sulfurtransferase (3MST) together with cysteine amino transferase (CAT), and D-amino acid oxidase (DAO) in concert with 3MST (1, 10, 37, 38). A detailed pathway of H2S synthesis has recently been reviewed by Beltowski (3). Although the distribution of H2S-producing enzymes is tissue specific, the kidney is known to express CBS and CSE (14). In diabetes, the level and activity of both these enzymes are reported to be impaired, and thus H2S production leads to endothelial and other cell type injury (18, 21, 44). A lower plasma H2S concentration in DN patients on hemodialysis was shown to correlate with the progression of atherosclerosis (23). Other researchers including our own study showed that H2S production in the diabetogen-induced (44, 49) or type-1 diabetic kidney (17, 18) is decreased.

Role of H2S in Diabetic Kidney Remodeling and Function

Excess matrix protein synthesis and deposition occur in the diabetic kidney. Recent reports suggest that the H2S-producing enzymes and its levels are decreased in diabetes. Upon endogenous induction or exogenous supplementation of H2S, matrix remodeling was mitigated, suggesting a correlation between H2S deficiency and matrix accumulation. In streptozotocin-induced diabetic rats, H2S therapy improved renal function, decreased glomerular basement thickening, mesangial expansion, and interstitial fibrosis (49). In the same study, the authors documented that the reduction of glucose-induced oxidative stress by H2S was mediated by activation of the Nrf2 antioxidant pathway that exerts an anti-inflammatory effect by inhibiting NF-κB signaling in in vitro podocytes (49). This finding is in line with our own study on the genetic type-1 diabetic model, where plasma H2S levels were low and associated with increased extracellular matrix deposition and reduced vascular compliance which was mitigated by H2S treatment (17). Previously, our laboratory demonstrated that matrix metalloproteinase-9 (MMP-9) diminishes H2S production in the
type-1 diabetic (Akita) model and this was, in part, by reducing CBS and CSE enzyme expression (18). Similar results were also observed in high-glucose treatment in in vitro experiments using glomerular endothelial cells (18). The regulation of CBS/CSE enzymes by MMP-9 was confirmed using Akita and MMP-9 knockout mice along with partial mitigation of renal remodeling in double knockout (DKO) mice (17). In addition, we demonstrated that H$_2$S therapy improves renal function (17).

**Signaling Cross Talk Between Gaseous Modulators in the Diabetic Kidney**

Recently, Lee et al. (20) reported that H$_2$S inhibited high glucose-induced protein synthesis in renal epithelial cells (21). In a separate study, the same group also reported tadalafil, a phosphodiesterase 5 inhibitor, abrogated high glucose-induced global protein synthesis and matrix protein laminin-$\gamma$ by increasing the expression and activity of H$_2$S-producing enzyme CSE (20). They also observed that in podocytes, tadalafil-induced AMP-activated protein kinase (AMPK) phosphorylation was mitigated by CSE inhibitor DL-propargylglycine and small interfering RNA against CSE in podocytes (20). The authors concluded that high-glucose-induced matrix protein synthesis in podocytes involved a complex interaction of the nitric oxide (NO)-H$_2$S-AMPK axis (20). To further define molecular mechanisms of H$_2$S signaling in diabetic conditions, we performed in vitro experiments using mouse glomerular endothelial cells. The cells were stimulated with high-glucose and treated without or with a H$_2$S donor. Our results suggested that high glucose induced markers of matrix accumulation and autophagy through a LKB1/STRAD/MO25 dependent pathway (Fig. 1) (16).

**The N-Methyl-D-Aspartate Receptor, H$_2$S, and Diabetic Nephropathy**

Although H$_2$S does not have a specific receptor, it has been reported to modulate the $N$-methyl-D-aspartate (NMDA) receptor in the brain (13). The NMDA receptor is a glutamate receptor and ion channel protein mainly found in the nerve cells involved in controlling synaptic plasticity and memory function (22). In 2002, Deng et al. (6) demonstrated that NMDA receptors were also expressed in the kidney cortex and exerted a tonic vasodilatory response. Inhibition of the receptors caused marked renal vasoconstriction and reduction in renal blood flow in response to glycine infusion (6). However, it was not known whether H$_2$S affects the NMDA receptor in the kidney to modulate function. For the first time, our laboratory demonstrated the role of H$_2$S in renal NMDA receptor-mediated remodeling and dysfunction (18). Our findings suggested that diabetic kidney remodeling was, in part, due to reduced H$_2$S production due to MMP-9-mediated inhibition of CBS and CSE enzyme activity. (18). This was associated with disruption of gap junction proteins, connexin-40 and -43. The above changes were reversed by exogenous H$_2$S supplementation (18). More recently, we demonstrated that an increased renal-resistive index (RI), excess ECM deposition, elevated plasma creatinine, and diminished renal vascular density and cortical blood flow were normalized with H$_2$S treatment in Akita kidneys (17). These findings suggest that H$_2$S has the potential to prevent diabetic nephropathy by preserving renal microvascular architecture and function. A possible pathway of H$_2$S-dependent renal remodeling and signaling mechanism is shown in Fig. 1.

**Perspective and Concluding Remarks**

In summary, studies on H$_2$S have revealed a significant role in a variety of physiological and pathophysiological processes. A decrease in H$_2$S level has been implicated in several pathologies, including diabetic nephropathy, in animals and humans (11, 47, 49). In contrast, increased H$_2$S levels have been reported to contribute to $\beta$-cell apoptosis, leading to reduced mass and thus insulin, neutrophil infiltration, and inflammation in lipopolysaccharide-induced endotoxic shock in the lung and liver and synovial inflammation (24, 39, 46). However, the measurement of H$_2$S in some of the earlier studies is inaccurate and overestimated due to the lack of sensitive techniques and partly due to its volatile nature. In addition, studies which showed improvement in pathology used crude H$_2$S donors such...
as sodium hydrogen sulfide (NaHS), which is not suitable for human therapy due to its short-time bioavailability and possible toxicity. Current research is now focused on developing novel H$_2$S-releasing compounds, such as GYY4137, AP39, and SG-1002, which are known to simulate the endogenous production of H$_2$S in the body. These compounds are safer than bolus doses of NaHS or sodium sulfide (Na$_2$S), which instantly releases H$_2$S. Furthermore, the slow-releasing compounds can deliver H$_2$S over a longer period of time. In animal models, GYY4137 has been shown to improve vasculopathy (26, 31).

Since supplementation of H$_2$S has the potential to prevent and/or improve DN by preserving renal microvasculature and function, additional experiments are needed to test the safety and efficacy using the newer H$_2$S-releasing compounds. In early phase I and phase II clinical trials, the novel H$_2$S donor SG-1002 significantly increased blood levels of H$_2$S and nitrite, suggesting increased nitric oxide availability in healthy and heart failure subjects (32, 33). To fully exploit the therapeutic potential of H$_2$S, larger multicenter cohort studies are required to investigate its beneficial effects not only in the diabetic kidney but also in the other organs affected by diabetes.

Recently, a clinical trial using N-acetyl cysteine (NAC), a derivative of cysteine and substrate for H$_2$S, has completed in patients with chronic kidney disease and patients on dialysis (ClinicalTrials.gov Identifier: NCT01232257). The objective of this study was to examine whether NAC treatment would increase plasma H$_2$S levels and decrease oxidative stress and inflammation. While the results are awaited, it can be speculated that the outcomes will likely provide us insight into the H$_2$S backup mechanism during nitric oxide deficiency in chronic kidney disease patients.

GRANTS

This work was supported in part by National Institutes of Health Grants HL-104103 and DK104653 and American Heart Association Award 155GDC2584013.

DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the authors.

AUTHOR CONTRIBUTIONS

Author contributions: U.S. provided conception and design of research; S.P. edited and revised manuscript.

No conflicts of interest, financial or otherwise, are declared by the authors.

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